

WHAT IS CLAIMED IS:

1. A method for screening and isolating transcriptional coregulatory proteins of transcription factors using a reverse yeast two hybrid system, comprising:

fusing a DNA encoding a first transcription factor or a fragment thereof containing a first transcriptional activation domain, which first transcription factor is not a glucocorticoid receptor, to a DNA encoding a second transcriptional activation domain to form a DNA encoding a first hybrid protein as bait on a first yeast expression vector, wherein the expression of the first hybrid protein formed of the first transcription factor or fragment thereof and the second transcriptional activation domain is under the control of a promoter which is inducible in a yeast host cell;

fusing a cDNA from a cell-specific or tissue-specific cDNA library to a DNA encoding a DNA binding domain of a second transcription factor to form a DNA encoding a second hybrid protein as prey on a second yeast expression vector for expression in a yeast host cell;

fusing a DNA encoding a reporter protein to a DNA containing a promoter and a DNA response element, which is the cognate DNA response element for the DNA binding domain of the second transcription factor, to form a reporter gene construct,

wherein the expression of the reporter protein is under the control of the promoter and the DNA response element;

transforming auxotrophic yeast host cells with the first yeast expression vector containing the DNA encoding the first hybrid protein as bait, the second yeast expression vector containing the DNA encoding the second hybrid protein as prey, and the reporter gene, together or separately in any order, to generate transformed yeast host cells, wherein the auxotrophic yeast host cells carry a DNA encoding a protein capable of overcoming the auxotrophy of the auxotrophic yeast host cells, the expression of which protein is controlled by a promoter and a DNA response element which is the cognate DNA response element for the DNA binding domain of the second transcription factor;

inducing the expression of the first hybrid protein in the transformed yeast host cells with an inducer;

first screening the transformed yeast host cells for the ability to grow on a culture medium lacking a growth-sustaining component required to complement or overcome the auxotrophy of the auxotrophic yeast host cells and for the ability to express the reporter protein;

screening transformed yeast host cells, which were observed in the first screening to have the ability to grow on a culture medium lacking a growth-sustaining component required to complement or overcome the auxotrophy of the auxotrophic yeast

host cells and the ability to express the reporter protein, for the inability to express the reporter protein in the absence of the inducer; and

isolating a transformed yeast host cell identified as being able to express the reporter protein in the presence of inducer but unable to express the receptor protein in the absence of inducer to further isolate a transcriptional coregulatory protein of the first transcription factor and/or its encoding DNA.

2. The method of claim 1, wherein the first transcription factor is a nuclear receptor.

3. The method of claim 2, wherein the nuclear receptor is a steroid receptor.

4. The method of claim 3, wherein the steroid receptor is human androgen receptor.

5. The method of claim 3, wherein the steroid receptor is human estrogen receptor alpha or beta.

6. The method of claim 1, wherein the first transcription factor is a transcription factor that is not a steroid or nuclear receptor.

7. The method of claim 1, wherein the promoter inducible in yeast is the galactose (Gal 1-10) promoter, which also has the property of being glucose-repressible.

8. The method of claim 7, wherein the inducer is galactose.

9. The method of claim 7, wherein said screening of transformed yeast host cells for the inability to express the reporter protein in the absence of inducer is performed in the presence of glucose.

10. The method of claim 1, wherein the reporter protein is β -galactosidase.

11. The method of claim 1, wherein the DNA response element is a LexA DNA response element.

12. The method of claim 1, wherein the auxotrophic yeast host cells are auxotrophic for Leu2.

13. The method of claim 12, wherein the protein capable of overcoming the auxotrophy of the auxotrophic yeast cells is Leu2.

14. An isolated androgen receptor transcriptional coregulatory protein which interacts with androgen receptor to regulate androgen-dependent gene expression, comprising:

(a) an amino acid sequence selected from the group consisting of SEQ ID NO:4, SEQ ID NO:6, SEQ ID NO:8, and SEQ ID NO:10;

(b) an amino acid sequence having at least 85% sequence identity to (a) and the activity of (a); or

(c) a fragment of (a) or (b) and having the activity of (a).

15. The protein of claim 14, which comprises the amino acid sequence of SEQ ID NO:4.

16. The protein of claim 14, which comprises the amino acid sequence of SEQ ID NO:6.

17. The protein of claim 14, which comprises the amino acid sequence of SEQ ID NO:8.

18. The protein of claim 14, which comprises the amino acid sequence of SEQ ID NO:10.

19. The protein of claim 14, which comprises an amino acid sequence having at least 85% sequence identity to (a) and the activity of (a).

20. The protein of claim 14, which comprises a fragment of (a) or (b) and having the activity of (a).

21. An isolated DNA molecule comprising a nucleotide sequence encoding the androgen receptor transcriptional coregulatory protein of claim 14.

22. The DNA molecule of claim 21, wherein said nucleotide sequence is selected from the group consisting of SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, and SEQ ID NO:9.

23. A self-replicable vector comprising the DNA molecule of claim 21.

24. A host cell transformed with the DNA molecule of claim 21.

25. A process for producing an androgen receptor transcriptional coregulatory protein which interacts with androgen receptor to regulate androgen-dependent gene expression, comprising:

cultivating the host cell of claim 24 to express and produce the androgen receptor transcriptional coregulatory protein; and

recovering the produced protein.

26. An antisense oligonucleotide complementary to a messenger RNA transcribed from the DNA molecule of claim 21, wherein said oligonucleotide inhibits the production of an androgen receptor transcriptional regulatory protein which interacts with androgen receptor to regulate androgen-dependent gene expression.

27. A molecule having the binding portion of an antibody capable of binding to the protein of claim 14.

28. The molecule of claim 27, which is a monoclonal antibody.

29. A method for treating an androgen dependent disease comprising administering to a patient in need thereof an effective amount of the molecule of claim 27.

30. The method of claim 29, wherein the androgen dependent disease is selected from the group consisting of prostate cancer, benign prostatic hyperplasia, and androgen-dependent hair loss.

31. A method of screening for and identifying inhibitors that disrupt the interaction between androgen receptor and androgen receptor transcriptional coregulatory protein, comprising:

providing an assay system for detecting and quantitating androgen receptor and androgen receptor transcriptional coregulatory protein interaction based on the level of activity of a reporter gene product produced upon interaction of the androgen receptor and the androgen receptor transcriptional coregulatory protein;

incubating androgen receptor and an androgen receptor transcriptional coregulatory protein with or without a potential inhibitor that disrupts the interaction between the androgen receptor and the androgen receptor transcriptional coregulatory protein, wherein the androgen receptor transcriptional coregulatory protein comprises an amino acid selected from the group consisting of SEQ ID NO:2, SEQ ID NO:4, SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, and SEQ ID NO:14;

determining the level of activity of a reporter gene product in the presence of the potential inhibitor relative to

the level of activity of the reporter gene product in the absence of the potential inhibitor;

identifying as an inhibitor any potential inhibitor for which said determining step determines that the level of activity of the reporter gene product in the presence of the potential inhibitor is substantially less than that in the absence of the potential inhibitor.

32. The method of claim 31, further comprising the step of isolating the inhibitor identified in said identifying step.

33. An inhibitor of androgen receptor and androgen receptor transcriptional coregulatory protein isolated by the method of claim 32.

34. A method for inhibiting the interaction between androgen receptor and androgen receptor transcriptional coregulatory protein, comprising contacting androgen receptor and androgen receptor transcriptional coregulatory protein with an inhibition effective amount of the inhibitor of claim 33.